Isolation and Characterization of *Bacillus anthracis-targeting* Phages as Potential Biodefense Agents

Sivan Alkalay-Oren, MSc[1], Ron Braunstein, MD[1], Shunit Coppenhagen-Glazer, PhD[1], Amit Rimon, MD, PhD[1,2,3], Ronen Hazan, PhD[1]

[1]Institute of Dental Science, Faculty of Dental Sciences, Hebrew University-Hadassah School of Dental Medicine, Jerusalem, Israel. [2]Department of Military Medicine and "Tzameret", Faculty of Medicine, Hebrew University of Jerusalem, Jerusalem, Israel. [3]Israel Defense Forces Medical Corps, Ramat Gan, Israel.

Abstract

Background: Anthrax remains a major biodefense threat because of the high mortality of inhalational disease, environmental persistence of spores, and the potential emergence of antibiotic-resistant strains. Bacteriophages represent a promising alternative antimicrobial strategy; however, only a limited number of *Bacillus anthracis-targeting* phages have been characterized.

Methods: We screened soil samples collected from multiple regions in Israel for lytic bacteriophages active against the avirulent *B. anthracis* Sterne strain using a modified agarose-top agar assay. We isolated and characterized three phages, designated Negev_SA, Carmel_SA, and Tavor_SA, by transmission electron microscopy, bacterial killing assays, whole-genome sequencing, and phylogenetic analysis.

Results: All three phages produced plaques on *B. anthracis* Sterne lawns – turbid for Negev_SA and clear for Carmel_SA and Tavor_SA – and showed antibacterial activity against logarithmic- and stationary-phase *B. anthracis* cultures. Carmel_SA and Tavor_SA reduced viable bacterial counts below the detection threshold at 18 h (>6-log colony-forming unit [CFU]/mL reduction versus untreated controls), whereas Negev_SA produced approximately 1-log and 3-log reductions in logarithmic and stationary-phase cultures, respectively. Electron microscopy revealed siphovirus-like morphology with hexagonal heads and long tails. Whole-genome sequencing identified linear double-stranded DNA genomes of approximately 40 kb with a G+C content of approximately 35%. Phylogenetic analysis demonstrated close relatedness to the known *B. anthracis* phages Gamma, Fah, and Wbeta within the class Caudoviricetes. Genome sequences were deposited in GenBank under accession numbers KY963369 (Tavor_SA), KY963370 (Negev_SA), and KY963371 (Carmel_SA).

Conclusions: These findings support further evaluation of these phages as candidate biodefense, decontamination, and adjunctive therapeutic agents against *B. anthracis*.

Keywords: Anthrax, Biological warfare, Public health, Bioterror, Bioassay.

Corresponding Author: Amit Rimon, MD, PhD - amit.rimon@mail.huji.ac.il - Israel Defense Forces Medical Corps, Ramat Gan, Israel

Submitted for publication: 10.2.2026
Approved for publication: 4.4.2026

To cite this article: Alkalay-Oren S, Braunstein R, Coppenhagen-Glazer S, et al. Isolation and characterization of Bacillus anthracis-targeting phages as potential biodefense agents. J Isr Mil Med June 2026; 22(70):12-22.

Introduction

Anthrax is a severe zoonotic disease caused by the spore-forming Gram-positive bacterium *Bacillus anthracis*. Its spores are environmentally stable, inhalational anthrax has high mortality, and B. anthracis has been used in biological warfare and bioterrorism. It therefore remains a major public health and biodefense concern [1]. In addition to its role as a potential bioterror agent, anthrax remains endemic in wildlife and livestock in several developing regions, where outbreaks can also spread to humans [2,3]. Current post-exposure management relies primarily on prolonged antibiotic treatment combined with vaccination [4,5]. However, concerns about delayed diagnosis, toxin-mediated disease progression, and the possible emergence of antibiotic-resistant strains or intentional exposure highlight the need for alternative antimicrobial strategies [1,5]. From a military medicine perspective, *B. anthracis* poses a substantial biosecurity threat that requires robust medical countermeasures. Potential exposure scenarios in military operations or biodefense incidents necessitate rapidly deployable therapeutic alternatives that can complement existing antibiotic-based protocols, particularly against antibiotic-resistant strains. Developing effective phage-based countermeasures is therefore a priority for military medical preparedness and operational biodefense strategies [2,3].

Bacteriophages (phages), viruses that specifically infect bacteria, have gained renewed attention as potential antibacterial agents [6,7]. Phages are specific bacterial predators that can target both planktonic and biofilm-associated bacteria [8,9]. In addition, phage therapy offers several advantages in biodefense settings, including rapid isolation of new phages against resistant strains and adaptability through co-evolution or engineering approaches [8,10]. Phage therapy currently lacks approval by the United States Food and Drug Administration because of persistent regulatory bottlenecks and a need for more definitive evidence from controlled clinical trials [11]. Although several *B. anthracis* - targeting phages have been described [3,11,12], the available collection remains limited. In military medicine and biodefense, expanding the repertoire of well-characterized, lytic *B. anthracis* phages is essential to develop robust countermeasures and ensure operational readiness against natural or intentional anthrax outbreaks.

Here, we describe the isolation and characterization of three phages that target *B. anthracis* in both logarithmic- and stationary-phase liquid cultures and on agar. We assessed their efficacy, sequenced their complete genomes, and compared them with the lytic phage Gamma [13].

Methods

Bacterial Strains and Culture Conditions

The primary bacterial strain used was *B. anthracis* Sterne. This strain is avirulent because it lacks the pXO2 plasmid and is capsule-deficient. Bacterial cultures were grown in Luria-Bertani (LB) broth (composed of 10 g/L tryptone, 5 g/L yeast extract, and 10 g/L NaCl, pH 7.0) or on LB agar plates (supplemented with 1.5% w/v agar). We defined exponentially growing (logarithmic phase) cultures by monitoring optical density at 600 nm (OD600) until it reached 0.4–0.6, corresponding to approximately 10^8 colony-forming units (CFU)/mL. We obtained stationary-phase cultures after overnight growth (16 h) in a 37°C shaker at 200 rpm.

This study involved no human participants or animals, so no IRB approval was required.

Soil Sampling and Site Selection

We collected soil samples from the Golan Heights. After collection, we stored soil samples at 4°C in sterile 50 mL polypropylene tubes for no longer than 48 h before downstream application to preserve microbial integrity. We then incubated 10 g of each soil sample in 50 mL of LB broth for 2 days at 37°C with shaking at 150 rpm before processing [14].

Phage Isolation and Screening

We isolated phages using a modified agarose-top agar method. We centrifuged soil suspensions at 2650 × g for 10 min, then filtered the supernatant through 0.22 µm pore-size filters. We screened the filtrate for non-growth zones (presumptive plaques) by adding 200 µL of bacterial culture to 3.5 mL of 0.5% agarose and pouring the mixture onto LB plates [15].

To differentiate true bacteriophages from non-specific antibacterial compounds, we picked presumptive growth inhibition zones from the agar and transferred them into sterile LB media. We incubated these samples for 24 h at 4°C to facilitate phage diffusion, then filtered and re-screened them using serial dilutions to confirm replicating phages by plaque formation. To ensure clonal purity, we picked individual well-isolated plaques with a sterile pipette tip and subjected them to three successive rounds of single-plaque purification. We suspended purified phage stocks in SM buffer (100 mM NaCl, 8 mM MgSO4, 50 mM Tris-HCl, pH 7.5) and stored them long-term at 4°C for active use or at -80°C in LB broth supplemented with 20% glycerol for archival storage.

PFU Count

Plaque-forming units (PFU) were counted using a modified agarose-top agar method. Briefly, exponentially growing bacterial cultures (200 µL of 10^8CFU/mL) were added to 3.5 mL of agarose (0.5%) and poured onto an LB plate. After drying, 10-fold serial dilutions of each phage sample were spotted (3 µL) on the bacterial lawn in triplicate, and the plates were incubated overnight at 37°C. PFU were calculated from plaque counts.

Phage Purification and Transmission Electron Microscope (TEM) Visualization

Confirmed phages, designated Negev_SA, Carmel_SA, and Tavor_SA, were purified from single well-isolated plaques. For morphological characterization, we prepared purified phage specimens using Gill's method. We visualized them using TEM (JEOL, TEM 1400 Plus) equipped with a charge-coupled device camera (Gatan Orius 600) [14].

Lytic Efficacy and Growth Kinetics

We assessed phage killing efficiency by monitoring the growth kinetics of *B. anthracis* Sterne. We performed experiments in 96-well plates using a plate reader (Synergy; BioTek) at 37°C with 5-second shaking every 20 min. Phages were added at a multiplicity of infection (MOI) of 1, corresponding to a bacterial concentration of 10^6 CFU/mL and a phage concentration of 10^6 PFU/mL, at time point 0 for logarithmic-phase cultures, and at a bacterial concentration of 10^8 CFU/mL and a phage concentration of 10^8 PFU/mL for stationary-phase cultures.

Quantification of Viable Bacteria (CFU Count)

At the 18-hour endpoint of the growth experiments, we quantified CFUs using 10-fold serial dilutions. We spotted each sample (5 µL) onto LB plates in triplicate and incubated them overnight at 37°C to determine the density of surviving bacteria [15].

Genomic Sequencing and Bioinformatic Analysis

We purified phage DNA using a phage DNA isolation kit (Norgen Biotek), prepared libraries with the Illumina Nextera XT DNA kit (Illumina, San Diego, CA, USA), and sequenced samples on the Illumina MiSeq platform. The quality of the 150-bp paired-end reads was assessed by FastQC (http://www.bioinformatics.bbsrc.ac.uk/projects/fastqc). We performed de novo assembly of end-trimmed reads using Geneious 11 and its plugins (Biomatters) [14].

We annotated the genome using the RAST server (http://rast.nmpdr.org/rast.cgi) [16]. We scanned the phages for resistance genes and virulence factors using Abricate (https://github.com/tseemann/abricate) with the following databases: NCBI, CARD, Resfinder, ARG-ANNOT, EcOH, MEGARES, PlasmidFinder, Ecoli_VF, and VFDB. Phage and prophage sequences were aligned using the Mauve and MAFFT plugins in Geneious 11 (Biomatters). Based on these alignments, we constructed a phylogenetic tree in Geneious Tree Builder using the Tamura-Nei model and the Neighbor-Joining method [14].

For analysis, we categorized the linear DNA genomes as "left" (L) or "right" (R).

Statistical Analysis

We performed all quantitative experiments, including bacterial growth kinetics and plaque-forming assays, in triplicate (n=3). Data are presented as the mean ± standard deviation (SD). We evaluated statistical significance between group means using a one-way

analysis of variance (ANOVA) followed by Tukey's honestly significant difference post hoc test for multiple comparisons. We performed statistical analyses in R (version 4.3.1) and visualized data in Microsoft Excel. Values of $P<0.05$ were considered statistically significant.

Results

Phage Isolation and Morphological Characterization

Screening of soil samples from diverse locations across Israel yielded three distinct bacteriophages active against B. anthracis Sterne. The isolated phages, designated Negev_SA, Carmel_SA, and Tavor_SA, produced plaques on bacterial lawns (Table 1, Figure 1a). Plaque sizes were 3.3, 2.8, and 2.2 mm for Negev_SA, Carmel_SA, and Tavor_SA, respectively; plaques were turbid for Negev_SA and clear for the other two phages (Table 1, Figure 1b). Transmission electron microscopy showed that the three phages were morphologically indistinguishable, with hexagonal heads and long tails (Table 1, Figure 1c).

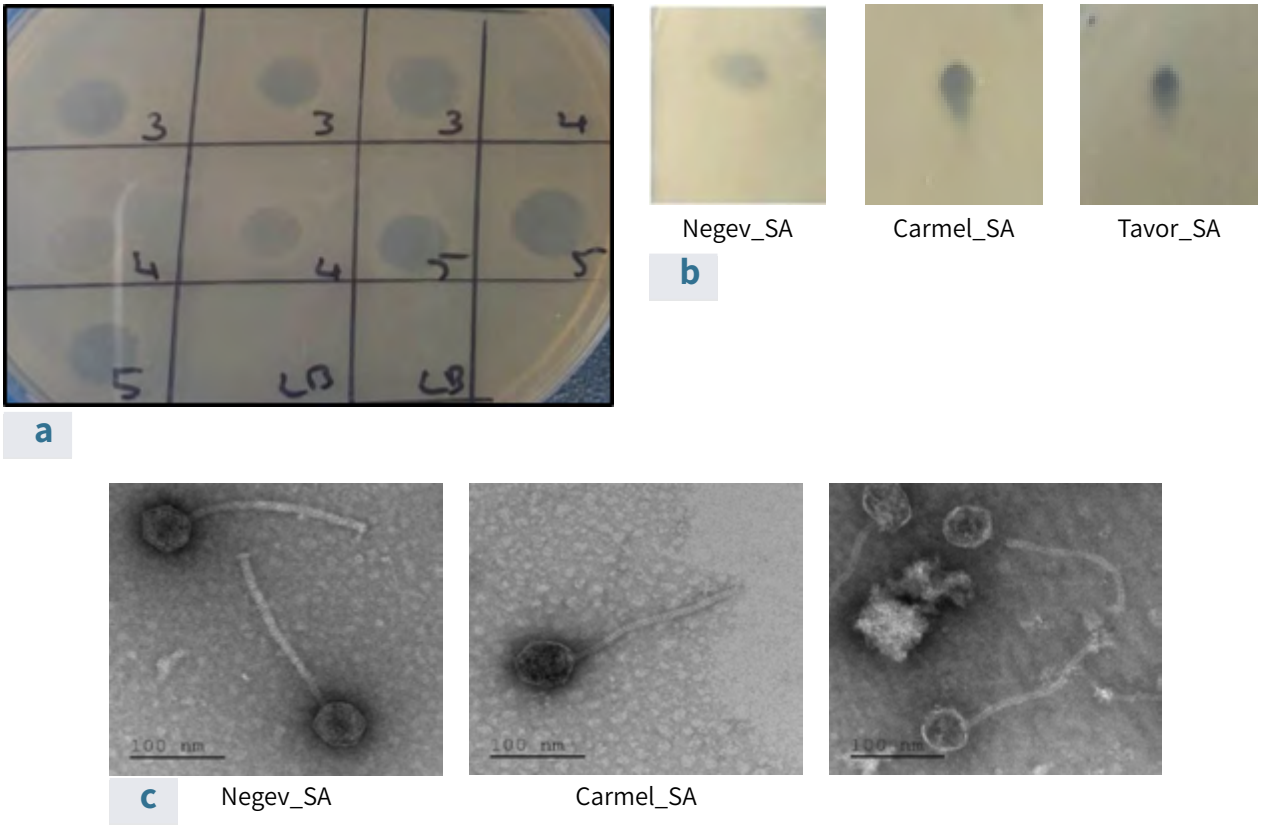


Figure 1. Isolation of Bacillus anthracis-targeting phages. (a) Environmental samples were screened for plaque formation on B. anthracis Sterne lawns, yielding three phages designated Negev_SA (3), Carmel_SA (4), and Tavor_SA (5). (b) Representative plaques formed by each phage on B. anthracis Sterne are shown. (c) Purified phage particles were visualized by transmission electron microscopy (TEM). Representative TEM images show the morphology of the isolated phages, including icosahedral capsids and long non-contractile tails characteristic of siphoviruses.

Characteristic	Negev_SA	Carmel_SA	Tavor_SA
ISOLATION AND MORPHOLOGY			
Host	*B. anthracis* Sterne		
Plaque morphology	Turbid	Clear	Clear
Plaque size (mm)	3.3	2.8	2.2
Morphology (TEM)	Siphovirus-like		
Capsid shape	Hexagonal (icosahedral)		
Tail type	Long, non-contractile		
Family	*Siphoviridae*		
Class	*Caudoviricetes*		
BACTERICIDAL EFFICACY (MOI = 1)			
Lytic activity (logarithmic and stationary phases)	Yes	Yes	Yes
CFU reduction — logarithmic phase (18 h)	~0.88 log	>6 log (below detection)	>6 log (below detection)
CFU reduction — stationary phase (18 h)	~3.59 log	>6 log (below detection)	>6 log (below detection)
Bacterial regrowth at ~10 h	Yes	No	No
Statistical significance (vs. untreated)	P<0.001	P<0.001	P<0.001
GENOMIC CHARACTERISTICS			
GenBank accession no.	KY963370	KY963371	KY963369
Genome type	Linear dsDNA		
Genome size (bp)	40 375	40 165	40 397
G+C content	34.9%	35.2%	35.2%
Unique genomic feature	Flagellar hook-length control protein (FliK)	β-galactosidase gene; phage terminase small subunit	~200 bp insertion in tail tape-measure protein

Table 1. Characteristics of the three isolated Bacillus anthracis-targeting phages.
Abbreviations: bp, base pairs; CFU, colony-forming unit; dsDNA, double-stranded DNA; MOI, multiplicity of infection; TEM, transmission electron microscopy. All killing assays were performed at MOI = 1 and analyzed by one-way ANOVA with Tukey's honestly significant difference post hoc test.

Bactericidal Efficacy Against B. anthracis

All three isolated phages showed lytic activity against B. anthracis Sterne in both logarithmic (Figure 2a) and stationary (Figure 2b) growth phases, with activity similar in magnitude to that of the reference phage Gamma [13]. Bacterial regrowth occurred after 10 h only in Negev_SA-infected cultures (Figure 2a). At the 18-hour endpoint, Carmel_SA and Tavor_SA reduced viable bacteria to below the limit of detection (P<0.001, Table 1, Figure 2c), whereas Negev_SA achieved a 0.88 log reduction in logarithmic-phase cultures and a 3.59 log reduction in stationary-phase cultures (P<0.001, Table 1, Figure 2c).

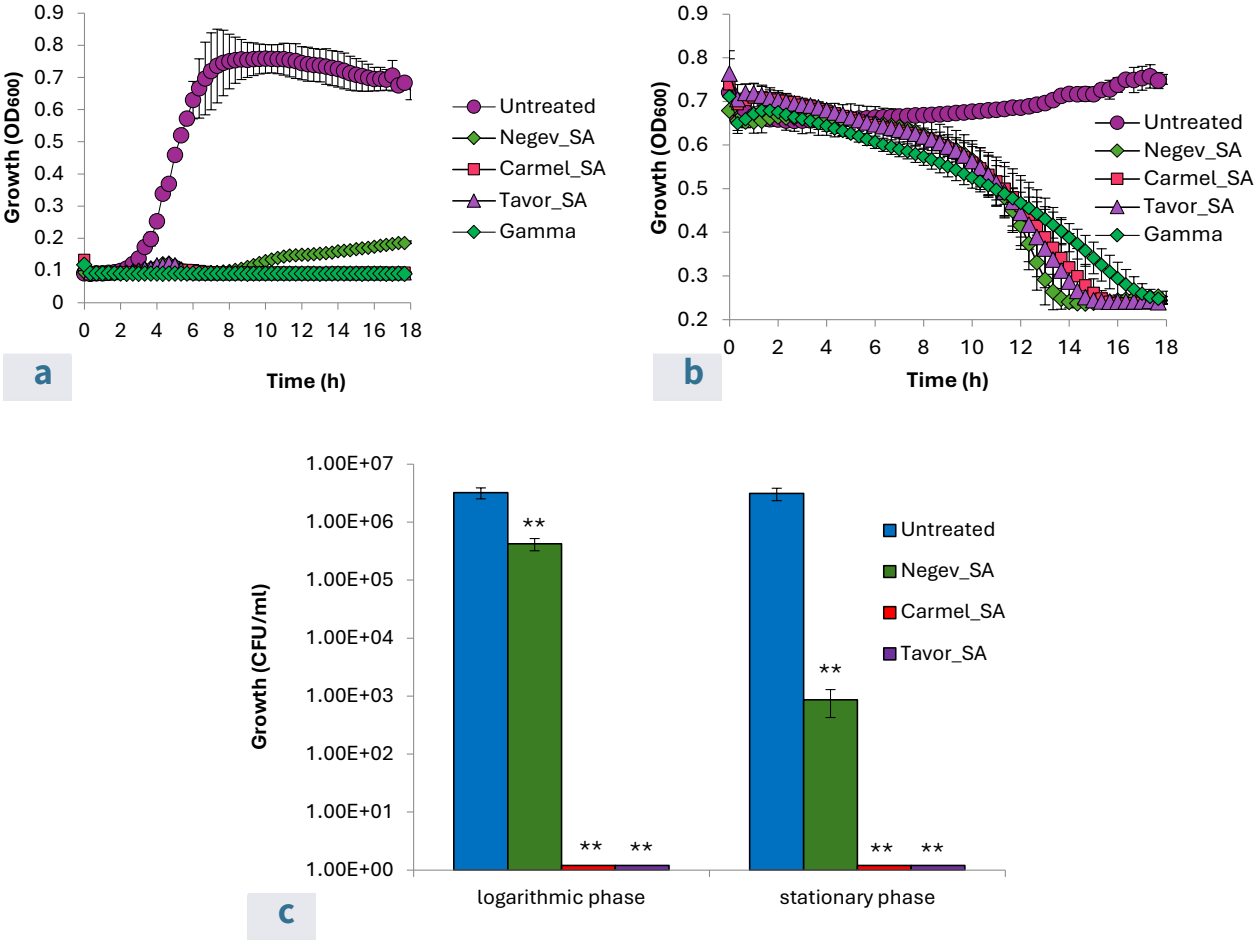


Figure 2. Efficacy of B. anthracis Sterne lysis by three new phages versus phage Gamma.
We compared the lytic effectiveness of the new phages with phage Gamma by measuring OD600 every 20 min in infected liquid cultures, with untreated cultures as controls.
(a) Growth curves of logarithmic-phase B. anthracis Sterne cultures following infection with Carmel_SA, Negev_SA, Tavor_SA, or Gamma at time point 0.
(b) Growth curves of stationary-phase B. anthracis Sterne cultures following phage addition at time point 0.
(c) Quantification of viable bacteria at the experimental endpoint (18 h) by colony-forming unit (CFU) enumeration. Data demonstrate differences in antibacterial efficacy among the isolated phages, with Negev_SA showing reduced suppression of bacterial regrowth compared with Carmel_SA and Tavor_SA.
Data are presented as the mean±SD of triplicate measurements. **P<0.001, determined by one-way ANOVA followed by Tukey's post hoc test.

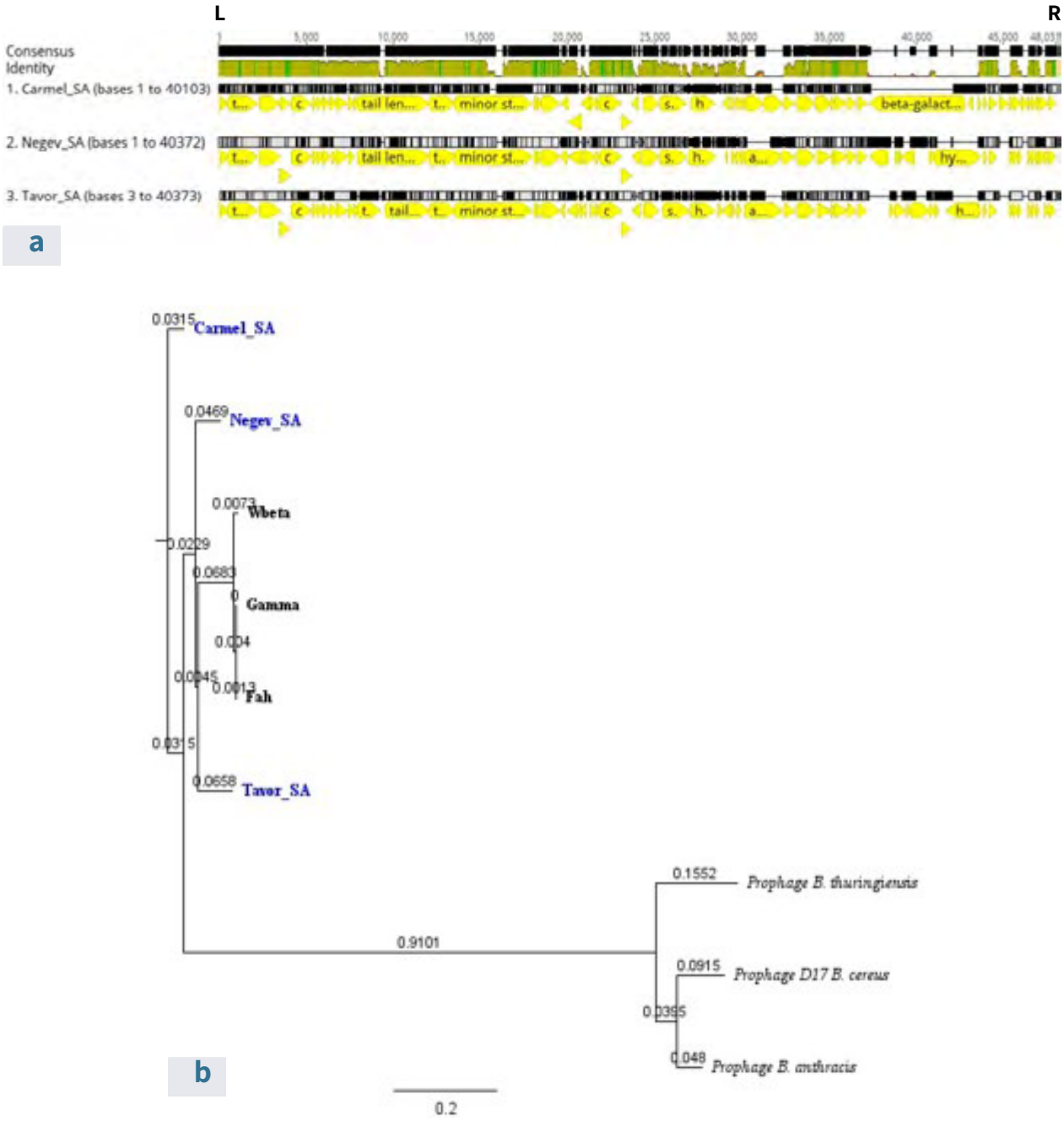


Figure 3. Genomic analysis of the B. anthracis-targeting phages. (a) Whole-genome alignment of Carmel_SA, Negev_SA, and Tavor_SA. Conserved genomic regions are indicated in green along the identity track. In contrast, regions with lower sequence similarity are shown in yellow or red, and gaps indicate absent or highly divergent regions. The left ("L") and right ("R") genome regions are indicated as described in the text. The left side of the genomes contains primarily conserved structural and replication-associated genes, whereas the right side contains more variable regions enriched in hypothetical proteins.
(b) Phylogenetic analysis of Carmel_SA, Negev_SA, and Tavor_SA based on whole-genome sequence similarity. The tree includes previously characterized B. anthracis phages and related Bacillus prophages showing the highest genomic similarity to the phages identified in this study, including Gamma, Fah, and Wbeta.

Genomic Characterization and Phylogenetic Analysis

Phage sequences are available in GenBank under accession numbers KY963369 (Tavor_SA), KY963370 (Negev_SA), and KY963371 (Carmel_SA, Table 1) [17].

The linear DNA genomes of Negev_SA, Carmel_SA, and Tavor_SA are similar in size (40 375 bp, 40 165 bp, and 40 397 bp, respectively), with a G+C content of approximately 34.9–35.2% (Table 1), similar to the *B. anthracis* genome (35.1%) [18]. The left side of the genomes is more conserved than the right side and contains structural and replication genes. In contrast, the right side contains mostly unannotated hypothetical genes with lower conservation. All three phages contain genes conserved among many phages, including an RNA polymerase sigma factor specific for sporulation, DNA replication proteins, holins, and numerous hypothetical proteins.

Several unique genomic features were identified. Negev_SA contains a flagellar hook-length control protein (FliK), Carmel_SA contains genes encoding β-galactosidase and a phage terminase small subunit, and Tavor_SA contains an insertion of approximately 200 bp in its tail length tape-measure protein, which is also present in several Bacillus prophages (Table 1).

All three phages contain lysogenicity-related genes, including site-specific recombinases, repressor, and anti-repressor genes. The repressor genes differ in length and sequence among Negev_SA, Carmel_SA, and Tavor_SA. We detected no known virulence factors or antibiotic resistance genes in the databases interrogated (see Methods).

Phylogenetic analysis showed that Carmel_SA, Tavor_SA, and Negev_SA cluster with other B. anthracis phages, including Gamma (NC_007458), Fah (DQ222851), and Wbeta (NC_007734.1), and show sequence similarity to prophages found in various Bacillus species (Figure 3a-3b).

Discussion

This work aimed to isolate phages against *B. anthracis*, with the long-term goal of evaluating their potential utility for biodefense applications and future therapeutic development. We isolated three phages displaying effective antibacterial activity against *B. anthracis* in both liquid cultures and plate-based assays. Their activity was similar in magnitude to that of the well-characterized phage Gamma, which has historically been used to identify *B. anthracis*.

From a military medicine and biodefense perspective, isolating three independently derived, genetically related phages with activity similar to that of the reference phage Gamma strengthens the foundation for developing a robust countermeasure arsenal. Multiple phages with similar lytic capabilities provide redundancy and mitigate the risk of bacterial resistance to a single phage variant, a critical consideration for military medical preparedness and post-exposure prophylaxis strategies.

In liquid culture experiments, Negev_SA showed lower antibacterial efficacy than Carmel_SA and Tavor_SA, as evidenced by increased bacterial regrowth and significantly higher CFU counts following infection. One possible explanation may relate to differences in the repressor genes identified among the three phages. Variability in repressor gene sequence and length could influence phage-host interactions or lysogenic behavior and may contribute to the phenotypic differences observed between the phages [19].

Although Carmel_SA and Tavor_SA formed clear plaques, which are typically associated with lytic activity, all three phages contain lysogenicity-associated genes, including recombinases, repressors, and anti-repressors. Similar observations have previously been reported for phage Gamma, which was originally isolated from lysogenic bacteria [13]. The presence of lysogenicity-related genes in the phages described here suggests possible temperate behavior under certain conditions. However, genomic evidence alone is insufficient to determine whether these phages can form stable lysogens in *B. anthracis*, and additional functional studies are required to establish their lysogenic potential. This distinction matters in therapeutic applications, as temperate phages are generally less desirable for phage therapy because of concerns about horizontal gene transfer and incomplete bacterial killing.

Phylogenetic analysis showed close relatedness among Carmel_SA, Tavor_SA, Negev_SA, and previously characterized B. anthracis phages, including Gamma, Fah, and Wbeta [13], supporting their classification

within the class Caudoviricetes with siphovirus morphology. Similarity to prophages found in diverse *Bacillus* species may reflect shared evolutionary origins and genetic exchange within the genus.

Expanding the collection of genetically diverse candidates is essential for mitigating resistance, a need highlighted by other recent isolation efforts [6,11,12], which also included phage-derived lysins [20]. For instance, Li et al. recently isolated two B. anthracis siphoviruses [11] that show genomic parallels to our isolates. Much like Tavor_SA, Carmel_SA, and Negev_SA, their phages have double-stranded DNA genomes of similar size (36.5 kb and 40 kb, compared with approximately 40 kb for ours). The lowest similarity was between Negev_SA and their A16R4 phage (81.56%), and the highest between Tavor_SA and A16R1 (94.81%). They maintain strong evolutionary ties to classic reference strains; while our phages cluster with Gamma, Fah, and Wbeta, their A16R1 isolate shares 99.98% sequence identity with the diagnostic phage AP631. However, a key functional divergence lies in their phenotypic applications. The phages characterized by Li et al. have explicit tail-associated exopolysaccharide depolymerases that form plaque halos, providing optimized host specificity for diagnostic identification. In contrast, our evaluation focused on long-term killing kinetics across different growth phases.

These findings also have potential implications for biodefense and clinical management of anthrax infections. Phage-based approaches could theoretically complement antibiotic therapy, particularly in scenarios involving antibiotic-resistant strains or environmental decontamination following accidental or intentional exposure events. However, several important translational considerations remain unresolved. Optimal dosing strategies, routes of administration, tissue penetration, and pharmacokinetic behavior of B. anthracis-targeting phages remain largely unknown. In addition, the emergence of bacterial resistance to individual phages may limit phage-based therapy and may necessitate phage cocktails or combination approaches with antibiotics. The observed genomic variability among the isolated phages may support future development of such combinatorial approaches.

This study has several limitations. The phages were evaluated as monotherapy without assessing potential synergy with antibiotics such as ciprofloxacin or doxycycline. Bacterial killing assays were conducted under only two growth conditions (logarithmic and stationary phase); additional stress conditions (biofilm formation, anaerobic environment) should be evaluated. The study lacks *in vivo* validation in animal models, which is essential before therapeutic advancement. The presence of lysogeny-associated genes warrants further investigation into potential temperate behavior. Screening for virulence factors and antimicrobial resistance genes was limited to the databases interrogated by Abricate and cannot exclude uncharacterized determinants; future characterizations should include targeted screening for toxin genes and functional safety testing. Two further limitations bear directly on the biodefense application proposed here. All experiments used *B. anthracis* Sterne, an avirulent, pXO2-negative, capsule-deficient strain; because the capsule is a plausible barrier to phage adsorption, activity against fully virulent encapsulated strains remains untested. All assays were also performed on vegetative cells in liquid culture, so activity against spores – the form that drives environmental persistence and the decontamination use we propose – was not assessed.

Contribution to Military Medicine

While Carmel_SA, Tavor_SA, and Negev_SA share genomic similarity to previously characterized phages (Gamma, Fah, Wbeta), their independent isolation from diverse Israeli geographic regions provides genetic redundancy and supports the availability of locally derived phage resources. This geographic and genetic diversity strengthens Israel's biodefense preparedness by providing multiple independent candidate agents that could be developed further or combined in phage cocktail approaches. Furthermore, their characterization under controlled laboratory conditions and comparative efficacy assessment support their further evaluation as candidates for biodefense countermeasure development.

Future directions

Future research should further investigate the biological significance of lysogenicity-associated genes in *B. anthracis*-targeting phages and determine whether these phages can form stable lysogens under clinically relevant conditions. Additional studies are also needed to evaluate phage efficacy in environmental decontamination settings, including contaminated surfaces and soil, which are relevant to anthrax outbreak control and biodefense preparedness.

Important areas for further study include characterizing phage pharmacodynamics and pharmacokinetics, optimizing dosing and delivery strategies, assessing resistance development during prolonged exposure, and evaluating efficacy and safety in relevant animal infection models [20]. Expanding the diversity of available *B. anthracis*-targeting phages and identifying obligately lytic candidates may further facilitate the development of robust phage-based countermeasures against future anthrax outbreaks or biosecurity threats.

Conclusions

The results presented here support continued investigation of Tavor_SA, Carmel_SA, Negev_SA, and related phages as potential biodefense tools against natural or intentional *B. anthracis* outbreaks. Phages offer several theoretical advantages, including high specificity, adaptability, and the potential to engineer or combine multiple phages to overcome bacterial resistance. Although substantial translational and safety challenges remain, *B. anthracis*-targeting phages may represent a promising complementary approach for future antimicrobial and environmental decontamination strategies.

Acknowledgments

The authors thank the Israel Ministry of Defense and the Rosetrees Trust for supporting this research. We thank Ida Steinberger-Levy from the Department of Biochemistry and Molecular Genetics, The Israel Institute for Biological Research, Ness-Ziona, Israel.

Author Contributions

RH designed the research. SAO, RB and SCG performed the experiments. SAO, AR and RB wrote the paper. All authors reviewed the manuscript.

Conflict of interest

The authors declare no conflicts of interest.

Source of Support

None to declare.

Disclaimer

The views expressed are the authors' own and not an official position of the institution, the funder, the IDF, HUJI or the IMA.

References

1. Sangwan N, Gangwal A, Jain P, et al. Anthrax: transmission, pathogenesis, prevention and treatment. Toxins (Basel) 2025; 17(2): 56.
2. Kracalik IT, Kenu E, Ayamdooh EN, et al. Modeling the environmental suitability of anthrax in Ghana and estimating populations at risk: implications for vaccination and control. PLoS Negl Trop Dis 2017; 11(10): e0005885.
3. Bozdeveci A, Karaoglu SA, Akman A, et al. Identification of the biological properties of three novel lytic bacteriophages capable of infecting Bacillus anthracis: novel members of the class Caudoviricetes. Vet J 2026; 316: 106598.
4. Manish M, Verma S, Kandari D, et al. Anthrax prevention through vaccine and post-exposure therapy. Expert Opin Biol Ther 2020; 20(12): 1405-1425.
5. Bower WA, Yu Y, Person MK, et al. CDC guidelines for the prevention and treatment of anthrax, 2023. MMWR Recomm Rep 2023; 72(RR-6): 1-47.
6. Gordillo Altamirano FL, Barr JJ. Phage therapy in the postantibiotic era. Clin Microbiol Rev 2019; 32(2): e00066-18.
7. Schooley RT, Biswas B, Gill JJ, et al. Development and use of personalized bacteriophage-based therapeutic cocktails to treat a patient with a disseminated resistant Acinetobacter baumannii infection. Antimicrob Agents Chemother 2017; 61(10): e00954-17.
8. Khalifa L, Brosh Y, Gelman D, et al. Targeting Enterococcus faecalis biofilms with phage therapy. Appl Environ Microbiol 2015; 81(8): 2696-2705.

9. Amankwah S, Abdella K, Kassa T. Bacterial biofilm destruction: a focused review on the recent use of phage-based strategies with other antibiofilm agents. Nanotechnol Sci Appl 2021; 14: 161-177.
10. Rimon A, Belin J, Yerushalmy O, et al. Pulsed blue light and phage therapy: a novel synergistic bactericide. Antibiotics (Basel) 2025; 14(5): 481.
11. Li L, Zhang H, Jin H, et al. Identification and characterization of two Bacillus anthracis bacteriophages. Arch Virol 2024; 169(7): 134.
12. Nakonieczna A, Rutyna P, Fedorowicz M, et al. Three novel bacteriophages, J5a, F16Ba, and z1a, specific for Bacillus anthracis, define a new clade of historical Wbeta phage relatives. Viruses 2022; 14(2): 213.
13. Schuch R, Fischetti VA. Detailed genomic analysis of the Wbeta and gamma phages infecting Bacillus anthracis: implications for evolution of environmental fitness and antibiotic resistance. J Bacteriol. 2006;188(8):3037-3051.
14. Rimon A, Yerushalmy O, Belin J, et al. Six novel Pseudomonas aeruginosa phages: genomic insights and therapeutic potential. Phage (New Rochelle) 2025; 6(1): 32-40.
15. Rimon A, Rakov C, Lerer V, et al. Topical phage therapy in a mouse model of Cutibacterium acnes-induced acne-like lesions. Nat Commun 2023; 14(1): 1005.
16. Aziz RK, Bartels D, Best AA, et al. The RAST server: rapid annotations using subsystems technology. BMC Genomics 2008; 9: 75.
17. Alkalay S, Sternberg S, Coppenhagen-Glazer S, Hazan R. Complete genome sequences of three Bacillus anthracis bacteriophages. Genome Announc 2018; 6(1): e01164-17.
18. Magome TG, Surleac M, Hassim A, et al. Decoding the anomalies: a genome-based analysis of Bacillus cereus group strains closely related to Bacillus anthracis. Front Microbiol 2025; 16: 1527049.
19. Howard-Varona C, Hargreaves KR, Abedon ST, Sullivan MB. Lysogeny in nature: mechanisms, impact and ecology of temperate phages. ISME J 2017; 11(7): 1511-1520.
20. Nakonieczna A, Topolska-Wos A, Lobocka M. New bacteriophage-derived lysins, LysJ and LysF, with the potential to control Bacillus anthracis. Appl Microbiol Biotechnol 2024; 108(1): 76.